Study of the carriage and characterization of a common bacterium, *Staphylococcus aureus*, on the campus of Concordia University St. Paul

Hannah Pedersen, Dr. Mandy Brosnahan,and
Dr. Taylor Mach

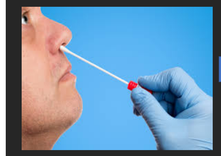
BACKGROUND: *Staphylococcus aureus* is a dual role bacterium and is able to live commensally in some patients, but can cause disease in others. Methicillin resistant *S. aureus* (MRSA) causes life threatening disease in patients and can be very hard to treat since it is resistant to many antibiotics. In order for *S. aureus* to wreak havoc on the body, it must be able to have specific genes expressed to secrete toxins. These toxins are what causes the patients to get a wide variety of symptoms like boils, scalded skin, toxic shock syndrome, pneumonia or sepsis. This study sets out to find the amount of people who carry *S. aureus* in their nasal cavity and if they do, is is MRSA and does it have the specific genes expressed to secrete toxins. As of 02/26/20, there have been 1065 swabs collected from the community of Concordia St. Paul's campus and 880 of those swabs have been processed through culture tests. Out of those 880 swabs, 234 have been positive for *S. aureus* and this is a 26.6% carriage rate.

TEST SIGNIFICANCES

- Mannitol Salt Agar (MSA) plate contains a high salt concentration and mimics what it would be like for a bacterium to grow on human skin. *S. aureus* can grow on this plate because it can thrive under high salt conditions. When it grows, it causes the fermentation of mannitol, which gives it the yellow color change on the plate *(2)*.
- Deoxyribonuclease (DNase) plate looks for the presence of DNase enzymes. The plate's medium contains DNA in it that can be broken down by a *S. aureus* enzyme called deoxyribonuclease, which you can see by the halo color change. The deoxyribonuclease catalyzes the hydrolytic cleavage of phosphodiester linkages in DNA backbone. This gives the bacteria nutrients and helps to support its growth *(3)*.
- Coagulase test is done to see if the bacterium has the coagulase enzyme, which will cause aggregation of red particles on the blue background. The coagulase enzyme converts fibrinogen to fibrin which gives *S.aureus* the ability to clot blood/plasma.Fibrin also can coat the bacteria and prevent it from getting phagocytosed *(4)*.
- Columbia Nalidixic Acid Blood agar (CNA) plate contains two antibiotics that doesn't allow gram negative organisms to grow on it. It also contains blood which allows for beta hemolysis to be seen. In order for the yellow glow to occur, the bacterium must possess hemolysins that cause blood cell lysis. *S.aureus* use their hemolysins to lyse RBC, which causes hemostatic disturbances *(1)*.
- Catalase test is looking for the presence of a catalase enzyme, and when it comes into contact with hydrogen peroxide, will cause O2 gas to be released (bubbles). *S.aureus* uses catalase enzyme to protect itself from oxidative damage (5).
- Gram stain is done in order to look at the bacterium under the microscope to make sure that it looks like a *S.aureus* bacterium. Typical *S.aureus* bacterium are Gram Positive (Purple color) and in cocci shape and grape like orientations.

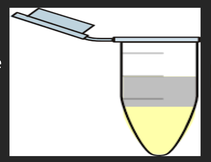
There is a 1 out of 4 chance that *Staphylococcus aureus* may be living in your nasal cavity right now.

Methods of Testing

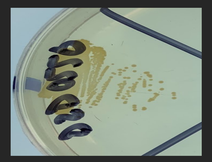


Nasal Swab

Isolate Swab

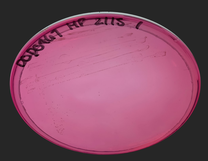


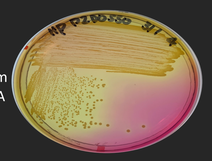
Grow isolated swab onto TSA Plate



Isolate bacterium onto MSA Plate

Negative MSA Plate: No yellow color change of medium surrounding colonies & smear of bacteria

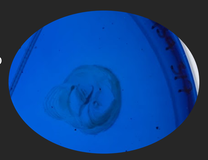


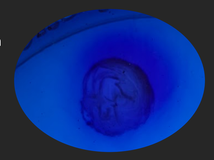


Positive MSA Plate: Yellow color change of medium surrounding colonies & smear of bacteria

Grow fresh bacterium on DNase plate

Negative DNase Test: no halo = negative reaction indicating no presence of DNase

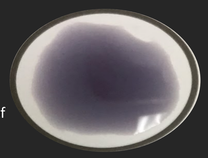


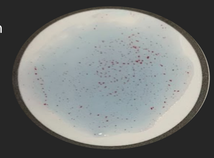


Positive DNase Test: halo seen = positive reaction indicating presence of DNase

Grow fresh bacteria for Coagulase Test

Negative Coagulase Test: no aggregation of particles (red particles on blue background), indicating presence of coagulase enzyme

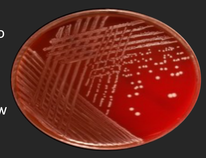


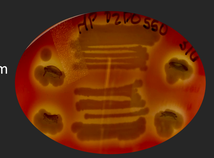


Positive Coagulase Test: aggregation of particles (red particles on blue background), indicating presence of coagulase enzyme

Grow bacterium on CNA plate

Negative CNA Test: No Alpha or Beta hemolysis due to presence of hemolysins= no yellow glow

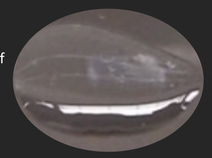


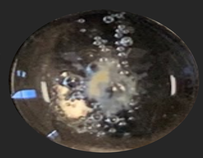


Positive CNA Test: Alpha or Beta hemolysis due to presence of hemolysins = yellow glow

Use bacterium to preform Catalase Test

Negative Catalase Test: no presence of catalase enzyme= no bubbles of O2 gas

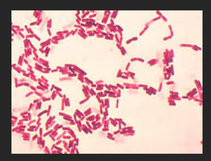


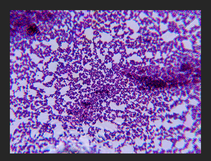


Positive Catalase Test: presence of catalase enzyme= bubbles of O2 gas

Do a Gram Stain on bacterium

Negative Gram Stain: Pink, Gram negative bacterium, in rods and chain orientation





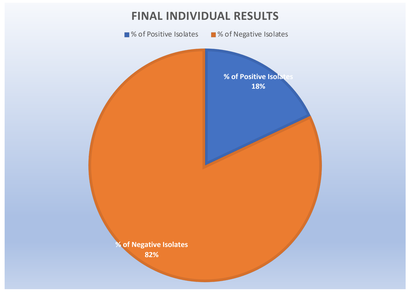
Positive Gram Stain: Purple, Gram positive bacterium in cocci/clusters

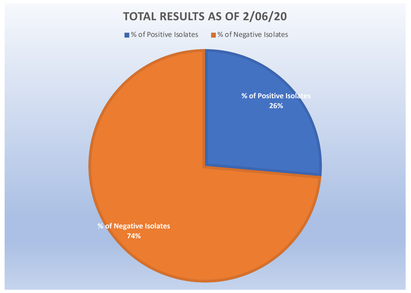
RESULTS

Isolate Tests

Isolate #'s	MSA	DNASE	Coagulase	CNA	Catalase	Gram Stain	Final Results
D2D0561	Negative	n/a	n/a	n/a	n/a	n/a	**Negative**
D2D0563	Positive	Negative	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Negative**
D2D0296	Positive	Positive	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Positive**
D2D0480	Negative	n/a	n/a	n/a	n/a	n/a	**Negative**
D2D0550	Positive	Positive	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Positive**
D2D0467	Negative	n/a	n/a	n/a	n/a	n/a	**Negative**
D2D0482	Positive	Negative	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Negative**
D2D0554	Negative	n/a	n/a	n/a	n/a	n/a	**Negative**
D2D0439	Positive	Negative	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Negative**
D2D0567	Positive	Negative	Positive	Positive	Positive	Gram +, purple cocci/cluster	**Negative**
D2D0569	Negative	n/a	n/a	n/a	n/a	n/a	**Negative**

There were eleven isolates that were tested in this study and out of the eleven, there were two that tested positive for being *S. Aureus.* This comes to a 18% carriage rate for these eleven isolates. For the study as the whole, as of 02/26/20 the carriage rate for positive isolates is 26.6%.





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5. K.Reiner, Catalase test protocol. *American Society for Microbio.* **1** (2010) [American Society for Microbiology].

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